Environmental Health Perspectives
Vol. 34, pp. 219-221, 1980

Influence of Post-Developmental Cadmium on Caries and Cariostasis by Fluoride

by T. R. Shearer,* J. L. Britton,* and D. J. DeSart*

Uptake of cadmium into molar enamel and dentin from rats receiving cadmium in their drinking water was markedly elevated and proportional to the amount of cadmium in the drinking water. Post-developmental cadmium did not influence caries development or alter the cariostatic effectiveness of fluoridated drinking water. The data indicate that the absolute concentration of cadmium in teeth is not predictive of cadmium-induced caries and that the critical period for caries promotion by cadmium may be during the developmental rather than the post-developmental period of tooth formation.

Introduction

Recent data obtained by using the rat caries model showed developmental cadmium to be strongly caries-promoting and to negate the cariostatic effects of fluoridated drinking water (*1*). Several publications (*2-5*) reported that post-developmental cadmium may also be caries promoting. Two of these studies (*2, 3*) used the older coarse corn diet, and the results of a third experiment (*4*) were marred because a nutritionally inferior diet was fed to the cadmium-treated animals. There are no data on the effect of post-developmental cadmium on cariostasis by fluoride. The purposes of the present research were: to re-examine the effect of post-developmental cadmium on caries and cariostasis by fluoride using the current standardized rat caries model and to study the uptake of post-developmental cadmium by enamel and dentin.

Materials and Methods

A total of 29, 14-day timed pregnant COB rats (Charles River, Wilmington, Massachusetts) were individually housed in plastic tubs with stainless steel tops containing cellulose bedding material (Bed O'Cobs, Anderson's, Maumee, Ohio). The animals received lab chow (Wayne-Lab-Blox, Allied Mills, Inc., Chicago, Illinois) and distilled water ad libitum. Two days after parturition, the pups were randomized and litters were culled to eight to ten pups. On days 19-21 the pups received daily oral inoculations (3 drops) of thioglycolate broth containing *Streptococcus mutans*-6715. On day 21, four or five pups of the same sex were placed in plastic tubs and fed cariogenic diet MIT-200 (Teklad, Madison, Wisconsin; 67% sucrose, 0.05 ppm cadmium, 0.5 ppm fluoride) and one of the following six drinking solutions ad libitum: distilled water (controls), 30 ppm Cd, 50 ppm Cd, 10 ppm F (fluoride controls), 10 ppm F + 30 ppm Cd, and 10 ppm F + 50 ppm Cd. Cadmium was present as $CdCl_2$ and fluoride was present as NaF. Animals were killed after 3 weeks, and caries in the first and second mandibular molars were scored by the method of Keyes (*7*). Caries data only for male rats are presented because female caries scores were too low to be useful.

The maxillary first and second molar teeth were extracted, dental pulp was removed from each with a burr, and enamel and dentin were separated by a single bromoform-acetone flotation method (*8*). Dental tissues and kidney samples were wet ashed in nitric acid (0.01*N*) and analyzed for cadmium by atomic absorption (Instrumentation Laboratories, Model 151, Wilmington, Massachusetts).

Differences between groups were tested for statistical significance by one-way analysis of variance followed by Neuman-Keul's post tests (*9*).

*University of Oregon Health Sciences Center, School of Dentistry, Division of Nutrition, Department of Biochemistry, Portland, Oregon 97201.

Table 1. Tissue uptake of post-developmental cadmium.

Diet group	Cd, ppm[a]		
	Enamel	Dentin	Kidney (wet)
Control	0.84 ± 0.08 (5)	0.55 ± 0.12 (5)	0.76 ± 0.12 (6)
30 ppm Cd	141.50 ± 29 (6)	30.30 ± 3.0 (6)	4.09 ± 0.42 (6)
50 ppm Cd	252.50 ± 44 (6)	53.90 ± 8.0 (6)	6.53 ± 0.76 (6)

[a]Mean ± S.E. (number of pools in parentheses).

Results

The addition of 30 and 50 ppm Cd to the drinking water caused a marked accumulation of cadmium into enamel and dentin in the molar teeth proportional to the amount of cadmium in the drinking water (Table 1). Cadmium accumulation was especially marked in enamel which rose from 0.84 ppm in the controls to 142 and 253 ppm Cd in the 30 and 50 ppm Cd treatment groups. Enamel accumulated more cadmium than dentin; the enamel to dentin cadmium ratio was 1.5 in controls, and this ratio increased to 4.7 in the two cadmium groups.

Levels of cadmium in the kidneys of animals receiving post-developmental cadmium increased with increasing amounts of cadmium in the drinking water up to 6.5 ppm in the kidneys of animals receiving 50 ppm Cd drinking water (Table 1). These soft tissue levels indicated appreciable absorption of dietary cadmium. When compared to the elevated cadmium levels in the teeth, they further emphasized the remarkable uptake of post-developmental cadmium in the enamel and dentin.

The effect of elevated doses of cadmium in the drinking water on dental caries is shown in Table 2. Post-developmental cadmium alone had no statistically significant effect on caries scores at either the 30 or 50 ppm level. This lack of effect of post-developmental cadmium on caries was evident in the total buccal caries scores, enamel scores and dentin slight scores. The levels of cadmium used in the drinking water in the present experiment covered the maximum physiological dose, since 30 ppm Cd alone caused a slight reduction in final body weights amounting to −6%, and 50 ppm Cd caused a −10% reduction in final body weights compared to controls.

Addition of 10 ppm fluoride to the drinking water of the rats in the present experiment was markedly cariostatic (Table 2). Fluoride alone reduced total buccal lesions by 94% compared to control rats. However, addition of 30 or 50 ppm Cd to the fluoridated drinking water had no consistent effect on the cariostatic properties of fluoride. The apparent increase in caries in the F + 30 ppm Cd group was believed to be spurious because of the low caries rates and the fact that this effect was not found in the F + 50 ppm Cd group.

Discussion

The high concentrations of cadmium found in the teeth of the animals in the present experiment were unexpected and are the highest ever reported for enamel tissue (*10, 11*). In swine consuming 1350 ppm Cd in the diet, whole incisors contained 212 ppm Cd (*11*). Literature values for the cadmium concentrations of teeth from man and experimental animals range from 0.5 to 10 ppm (*10, 11*). Bawden and

Table 2. Effect of post-developmental cadmium on caries in male rats.

Drinking water group	n	Number of lesions			Body weight, g
		Total buccal	Enamel	Dentin slight	
Control	18	21.9 ± 3.5	11.7 ± 1.2	6.7 ± 1.3	175 ± 4
30 ppm Cd	21	16.2 ± 3.5	10.0 ± 1.4	4.0 ± 1.3	164 ± 3[b]
50 ppm Cd	21	15.4 ± 2.8	10.1 ± 1.1	3.5 ± 1.0	158 ± 4[b]
F Control	17	1.4 ± 0.4	1.4 ± 0.4	0.0 ± 0.0	159 ± 5
F + 30 ppm Cd	21	6.2 ± 1.1[c]	5.5 ± 1.0[b]	0.3 ± 0.2	167 ± 3
F + 50 ppm Cd	21	3.0 ± 0.6	3.0 ± 0.6	0.0 ± 0.0	162 ± 4

[a]Mean ± S.E.
[b]Means within the same vertical column were statistically different from the appropriate control group.
[c]Statistically different from F + 50 ppm Cd group.

Hammarstrom (*12*) found the enamel organ of the developing rat molar accumulated more radioactive cadmium than all other hard tissues. In an experiment where cadmium was injected directly into lactating pups, the highest treatment groups accumulated 15.8 ppm Cd in the developing enamel (*1*). In bovine and human teeth containing approximately 1 ppm Cd, there was no accumulation of cadmium with age indicating that under conditions of normal cadmium exposure, most of the cadmium in teeth was deposited during tooth development (13). Cadmium exposure was started in the present experiment on day 21 post partum; at this time crown formation had been completed and tips of the crowns had just erupted into the oral cavity. Thus, the enamel underwent post-eruptive maturation during the period of cadmium administration. This phase of enamel development may be responsible for the excessive uptake of cadmium in the molar teeth in the present experiment. Nothing is known about the anatomical location of cadmium in teeth. In fluorotic incisor teeth, fluoride is excessively high in hyperplastic pits containing coronal cementum (*14*). Similar experiments should now be performed to test if excessive post-developmental cadmium may likewise be located in specific regions of enamel.

Despite the marked uptake of post-developmental cadmium into dental tissues, post-developmental cadmium did not appreciably influence caries development or alter the cariostatic properties of fluoride. This is in contrast to our previous publications showing a strong caries promoting effect of developmental cadmium, which also partially negated the cariostasis of simultaneously administered fluoride (*1*). Injections of 0.75 mg Cd per kg body weight during lactation increased caries by 183% and reduced the effectiveness of fluoride by 40%. Although in the present experiment administration of cadmium was by oral route rather than injection as in previous experiments, our results confirm the concept that exposure to cadmium during the period of tooth development is the critical time for cadmium-induced caries as opposed to post-developmental cadmium.

In developing teeth, the uptake of cadmium by enamel was directly related to an increased frequency of caries in female rats (*1*). Enamel cadmium levels resulting from cadmium administration during tooth development were much lower (< 2.0 ppm) than in the present post-developmental experiment (~253). Although Curzon and Crocker (*15*) found evidence for a mathematical association between increased levels of cadmium in human enamel and caries, our results indicate that the absolute concentration of cadmium in enamel may not necessarily be predictive of cadmium-induced dental caries. In soft tissues much of the cadmium is associated with the protein fraction (*16*). As has been done with other trace elements (*17, 18*) experiments should now be performed to learn the chemical form of cadmium in enamel. It is possible that there may be more than one chemical form of cadmium in enamel, and one of these, possibly a protein-bound cadmium, may increase the susceptibility of enamel to caries.

This investigation was supported, in part, by U.S.P.H.S. Grant DE-04616 from the National Institute of Dental Research, National Institutes of Health, Bethesda, Maryland.

REFERENCES

1. Shearer, T. R., Britton, J. L., DeSart, D. J., and Johnson, J. R. Influence of cadmium on caries and the cariostatic properties of fluoride in rats. J. Dent. Res., submitted.
2. Ginn, J. J. and Volker, J. F. Effect of cadmium and fluorine on rat dentition. Proc. Soc. Exptl. Biol. Med. 59: 189 (1944).
3. Leichester, H. M. The effect of cadmium on the production of caries in the rat. J. Dent. Res. 25: 337 (1946).
4. Tamura, T., and Moriya, Y. Studies on the toxicity of drugs (XI). Toxicity of cadmium (abstract). Japan. J. Pharm. 23: 227 (1973).
5. Wisotzky, J., and Hein, J. W. Effects of drinking solution containing metallic ions above and below hydrogen in the electromotive series on dental caries in the Syrian hamster. J. Am. Dent. Assoc. 57: 797 (1958).
6. Navia, J. M. Experimental dental caries. In: Animal Models in Dental Research, University of Alabama Press, Birmingham, 1977, Chapt. 13.
7. Keyes, P. H. Dental caries in the molar teeth of rats. II. A method for diagnosing and scoring several types of lesions simultaneously. J. Dent. Res. 37: 1088 (1958).
8. Manley, R. S., and Hodge, H. C. Density and refractive index studies of dental hard tissues. I. Methods of separation and determination of purity. J. Dent. Res. 18: 133 (1939).
9. Bruning, J. L., and Kintz, B. L. Computational Handbook of Statistics. Scott Foresman and Company, Glenview, Ill., 1977.
10. Losee, F. L., Cutress, T. W., and Brown, R. Natural elements of the periodic table in human dental enamel. Caries Res. 8: 123 (1974).
11. Cousins, R. J., Barber, A. K., and Trout, J. R. Cadmium toxicity in growing swine. J. Nutr. 103: 964 (1973).
12. Bawden, J. W., and Hammarstrom, L. E. Distribution of cadmium in developing teeth and bone of young rats. Scand. J. Dent. Res. 83: 179 (1975).
13. Shearer, T. R., Johnson, J. R., and DeSart, D. J. Cadmium gradient in human and bovine enamel. J. Dent. Res., in press.
14. Shearer, T. R., Kolstad, D. L., and Suttie, J. W. Electron probe microanalysis of fluorotic bovine teeth. Am. J. Vet. Res. 39: 1393 (1978).
15. Curzon, M. E. J., and Crocker, D. C. Relationships of trace elements in human tooth enamel to dental caries. Arch. Oral Biol. 23: 647 (1978).
16. Friberg, L., Piscator, M., Nordberg, G. F., and Kjellstrom, T. Transport, distribution, and excretion of cadmium in animals. In: Cadmium in the Environment, CRC Press, Cleveland, 1974.
17. Shearer, T. R. Developmental and post-developmental uptake of dietary organic and inorganic selenium into the molar teeth of rats. J. Nutr. 105: 338 (1975).
18. Bibby, B. G., and Little, M. F. Fluoride and titanium and the organic material of dental enamel. J. Dent. Res. (Spec. Iss. B) 54: 131 (1975).